



شرکت تحقیقاتی تولیدی پویا ژن آزما

POUYA GENE AZMA Co. (PGA)

PGA Bacterial DNA Extraction kit

CatalogNo.PD115-050Quantity: 50 Preps

Store: RT

KIT CONTENTS:

Buffer I	5.5 ml
Buffer II	11 ml
Buffer III	8 ml
Buffer X	10 ml
Rnase A	50 µl
Solvent Buffer	6 ml

Need contents:

cool ethanol %100
cool ethanol %70

Attention: before use , please add RNase A to Buffer I and keep in 4 °C

LABORATORY PROTOCOL:

- 1: Collect 3 -5 ml bacterial cultures by centrifugation 13000 rpm for 3 m in microtube.
- 2: Resuspending of precipitate in 100µl Buffer I.
- 3: Add 200 µl Buffer II in tube and gently inverting 3-5 times.
- 4: Add 150 µl Buffer III in tube and gently inverting 3 times and keep in 37 °C for 45 minutes. (for gram negative bacteria no need number 4 step)
- 5: Add 180 µl Buffer X in tube and inverting for 10 times. (when add MIII buffer in tube and inverting, producing white precipitate in microtube)
- 6: Microcentrifuge 13000 rpm for 10 minutes.
- 7: Transfer of supernatant in new tube. (important: do not transfer of precipitate in new tube. If transfer of precipitate in it please repeat 9 and 10 steps again)
- 8: Add 2 volumes cool ethanol %96 - %100 in solution and gently inverting for 5 times.
- 9: Microcentrifuge 13000 rpm for 5 minutes.
- 10: Pour off the ethanol by gently inverting of tube and keep precipitate.
- 11: Washing the precipitate by adding 700µl cool ethanol %70 and inverting 2-3 times.
- 12: Microcentrifuge 13000 rpm for 1 minute.
- 13: Pour off the ethanol completely and dry pellet for 1-2 minutes in room temperature.
- 14: According to precipitate add 20- 50 µl Solvent Buffer in tube. The precipitate must to be solve completely.